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OCCURRENCE OF NEOPLASMS IN THREE RELATED INDIVIDUALS OF LABORATORY-BRED *PEROMYSCUS*

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INTRODUCTION

ALTHOUGH neoplasms and their mode of inheritance have been widely studied in various strains of the house-mouse (*Mus musculus*), we know of no previous records of such growths in *Peromyscus*. In the Laboratory of Vertebrate Biology, where some eighty thousand *Peromyscus* have been reared, tumors have occurred only rarely, and the ones previously examined were probably the result of infections. It is, therefore, of interest to record three recent occurrences of neoplasms in this genus of mice. *Peromyscus* has, in the laboratory, been exceptionally free from diseases of all kinds.

The individual mice in which the tumors developed were fed the usual balanced ration described by Dice (1934), except that hemp seed and canary seed were omitted from the formula and calcium carbonate was added. In this and other respects they received the same care accorded all the mice in the laboratory (Dice, 1929). It should be emphasized that clean cages were provided for the mice every month. The temperature of the mouse rooms is kept at approximately 72°F. during the winter months, but it rises somewhat beyond this during the summer months.

The senior author removed the tumors and studied their pathology and the junior author provided the pedigree data and life history. We are indebted to Lee R. Dice for suggesting the study and for much helpful advice.

PATHOLOGY

The tumors of the three affected mice will be described in sequence. Dr. A. F. French, of the Pathology Department, University of Michigan, confirmed the pathological diagnoses.

Mouse No. 72316, Laboratory of Vertebrate Biology; specimen No. 4484, pathology collection, Department of Dermatology and Syphilology. A hybrid male of the *Peromyscus maniculatus* group.

The tumor was in the left flank region of this mouse and was first noted during the latter part of December, 1946. At that time the mouse was approximately 184 weeks old, which is an advanced age for specimens of this genus. At the time of operation, March 18, 1947, the mouse had attained an age of 195 weeks (three and three-fourths years).

On gross inspection this mouse appeared mildly, chronically debilitated, but not acutely ill. His abnormality was centered around a large tumor mass bulging from his left flank region, dragging on the ground and impeding his progress in a mechanical way. Inspection of the tumor mass revealed multiple, small, superficial erosions over the area which came into contact with the ground as the mouse dragged itself along. The tumor fairly well covered the area of the left external oblique muscle. Palpation in the unanesthetized state revealed no apparent tenderness nor pain on the part of the animal, and the mass, which had the consistency of a tennis ball, was quite firm. It was securely attached to the overlying skin but was completely free of all attachments to the underlying structures. In shape it was roughly spherical, slightly lobulated, and three centimeters in diameter. During a period of about three months, which elapsed between discovery and excision, the tumor about doubled in apparent bulk.

Under light anesthesia the skin over this mass was incised, and the entire tumor mass was shelled out with a minimal amount of difficulty after separating the layers of the abdominal musculature and releasing a few adhesions. The abdominal and thoracic cavities were then opened, and the viscera contained therein were examined. Grossly, they appeared perfectly normal in size, shape, position, and functioning. It was our impression that if this operation had been done under sterile conditions, the mouse would have survived.

The excised tumor displayed virtually the same characteristics noted above. Its outer core was hard and firm, approximately one centimeter in thickness and dull white and lusterless. The central part was hollow and contained two or three cubic centimeters of thick, greenish, purulent material.

The first impression on gross examination was that this was a benign connective-tissue tumor, probably a fibroma, but rapid growth, as evidenced by the central caseation and necrosis, favored its diagnosis as a malignant process.

Parts of the tumor were preserved in alcohol and later sectioned and stained with hematoxylin and eosin. With reversed ocular the stained slide showed what appeared to be a fairly well-ordered arrangement of tissue peripherally, and a heterogeneous, ill-defined mass of tissue centrally. Under the low power (100×) the tumor was observed to be formed of fibrous connective tissue cells, arranged in a rather disorderly manner with many well defined connective tissue fibers running between them. Under high dry power (450×) many of these cells were seen to be undergoing mitotic division. Toward the center of the mass, seen under low power but much more clearly under high power, there were areas of white blood cell infiltration and loss of cellular definition and structure. The connective-tissue fiber strands were broken off and degenerated. There was some evidence of tissue compactness and compression. No tubercles were present. This region of the section shows definite necrosis of the tissue.

The neoplasm gave the impression of being a fibrosarcoma. It was apparently a rapidly growing one, which was only locally malignant and which showed no tendency to metastasize.

Mouse No. 76002, Laboratory of Vertebrate Biology; specimen No. 4499, pathology collection, Department of Dermatology and Syphilology. A male *Peromyscus maniculatus blandus,* from the Tularosa (New Mexico) stock.

The neoplasm was in the right flank region of this mouse and was first noted on March 26, 1947, when the mouse was 151 weeks old. At the time of operation, May 1, 1947, the mouse had attained an age of 156 weeks (three years).

On gross inspection this mouse appeared thin, emaciated, and chronically debilitated. He did not seem acutely ill, but due to a huge tumor mass bulging from the right flank, his movements were impeded much as in the animal described above.

The external surface of the tumor was completely eroded, leaving what appeared to be a single large craterlike ulcer with firm, raised edges and a deep punched-out center which was filled with foul-smelling caseated material. The tumor mass covered the entire left lower quadrant of the abdomen. The outer shell was firm and was securely attached to the surrounding skin, but completely free of all apparent attachments to the underlying structures.

Under ether anesthesia the skin surrounding the mass was incised, and the entire tumor mass, which rested on but was not attached to the peritoneum, was shelled out as in the former specimen. Unfortunately, at this time the mouse began to recover from the anesthesia and, following reanesthetization, it died. Gross examination of the abdominal and thoracic viscera revealed apparent normality in size, shape, position, and functioning. If the mouse had not been so markedly emaciated and had been able to withstand the ether anesthesia, it also might have survived the operation.

The excised tumor grossly bore out the characteristics noted above. The hollow hemispherical mass had a thin, firm outer core which was four to six millimeters in width and was dull, light, and lusterless. The central part of this hollow hemisphere was filled with thick material characteristic of caseation necrosis.

In view of the recent experience with mouse No. 72316, it was thought probable that this also was a fibrosarcoma which had grown rapidly and by virtue of its growth and caseation had resulted in the extreme emaciation of the mouse. Under microscopic examination the sections of the tumor showed the same structure as that of the previously described specimen and thus confirmed this opinion.

Mouse No. 82106, Laboratory of Vertebrate Biology; specimen No. 4642, pathology collection, Department of Dermatology and Syphilology. A female *Peromyscus maniculatus blandus* from the Tularosa (New Mexico) stock.

The tumor was first noted on February 6, 1948, and was removed on February 10, 1948, when the mouse was approximately 112 weeks (two years) old.

The tumor was on the right lateral chest wall just posterior to the right axilla in the anterior axillary line, where it was attached only to the skin. It was spherical, firm, and approximately two and one-half centimeters in diameter. There were two tiny ulcerated areas on its surface where this dragged along the ground as the mouse moved about.

Removal of the tumor under ether anesthesia confirmed the above findings, and the entire mass was easily shelled out through a simple skin incision. In the bisected tumor a firm wall, approximately one-half to three-fourths centimeter thick, encircled a hollow center which was filled with thick muco-serous material in contrast to the cheesy caseation in the two tumors previously described.

Examination of the mouse revealed no other gross abnormalities.

Under the reversed ocular a section of this mouse tumor had much the same general appearance as the other two, namely, an outer zone of cellular elements and a central area which appeared quite homogeneous, the caseated center of the tumor.

Microscopic examinations confirmed the caseation centrally, but peripherally the zone of cellular material showed none of the regularity of structure noted in the previously described tumors. Instead, there was a heterogeneous, irregular, disorderly arrangement of connective tissue and glandular elements, with small areas of necrosis scattered through them. We considered a diagnosis of adeno-carcinoma, but the findings were so poorly defined that the final diagnosis was "ill defined, malignant neoplasm of glandular and connective tissue with caseation." Again, the malignancy appeared only local in character.

HEREDITY OF THE AFFECTED ANIMALS

Certain ancestors are common to the three mice that developed the above described tumors.

Mouse No. 72316 was a hybrid produced by crossing four subspecies of *Peromyscus* belonging to the *maniculatus* group: *Peromyscus polionotus albifrons* of a stock originally taken in Gulf State Park, Alabama, and called "Gulf Shores II" in the records of the Laboratory of Vertebrate Biology; *P. leucocephalus* from Santa Rosa Island, Florida, called "Navarre"; *bairdii* from Allegan County, Michigan, called "Todd Farm"; and *blandus* from Tularosa, New Mexico. The original cross was between *albifrons* and *leucocephalus*. Later, *bairdii* and *blandus* of the black-tipped gray, light selection line (Blair, 1947) were bred into the line. A hybrid of this descent was the male parent and *blandus* (black-tipped gray, dark selection line) the female parent of the present individual. There was no immediate inbreeding between these two parental strains and only a moderate amount within the pedigree. In the fifth previous generation twenty-one different ancestors appear instead of the thirty-two required for a noninbred stock.

At an age of 138 weeks (two and two-thirds years) this mouse was mated for six months (April to October, 1946) to a sexually mature male of a similar hybrid line, but no offspring were produced. No pathologic growths were recorded for the parents, for fifty-four parental sibs, or for fifteen sibs of this mouse. None of these relatives, however, remain in the laboratory stock, and none were specially examined for neoplasms.

Mouse No. 76002 was from the brown-tipped buff strain of *P. m. blandus* (Blair, 1947). Its parents were sibs, and the same three pairs of field-caught animals appear in its fifth, sixth, and seventh ancestral generations. Thus, there was a considerable amount of inbreeding in the line. There were only twelve different ancestors in the fifth previous generation instead of the expected thirty-two.

The mouse was mated to a sexually mature female of the same strain and at an age of about 109 weeks (two years) a litter was produced. The mating continued in effect until April, 1947, during which period the female bore ten litters totaling twenty offspring. Three offspring are still alive. They have reached approximate ages of two and one-half years, but have no external signs of tumors. Six others examined when they had attained ages of one year and four months to two years also showed no external signs of tumors. No records of growths appear on the cards for the remaining offspring, all of which are now dead. Also, there are no records of growths for ten of the sibs of mouse No. 76002 or for its parents or their sixteen sibs.

Mouse No. 82106 was a female *P. m. blandus* of the brown-tipped gray, selection line (Blair, 1947). Two grandparents and two great grandparents of this mouse were litter mates. Thus, a moderate amount of inbreeding had occurred in its direct ancestors. Three sibs of No. 82106 are still alive in the laboratory. At an age of a little over two years neither shows any external signs of abnormal growths. There are no records of abnormal growths for twenty-four sibs that are no longer alive or for the parents or their twelve sibs.

At an age of seven months this mouse was mated to a male of the same strain, and twenty-three offspring were produced during a period of fifteen months (July, 1946, to November, 1947). Thirteen of these offspring are still alive and have reached ages from about one and one-half years to less than a year. None of them has any external signs of tumors. Ten of the offspring which are no longer alive had no records of tumors, but none of them had reached two years of age when they were removed from the stock.

Two of the mice, Nos. 72316 and 76002, which developed fibrosarcomas, had certain *blandus* ancestors in common. One pair of brown-tipped buff mice (female No. 49979 and male No. 51529) appears in the seventh ancestral generation of mouse No. 72316 and in the fifth ancestral generation of No. 76002. This pair from the second laboratory-bred generation was the offspring of female No. 43202 and male No. 43704 (first laboratory-bred generation), which appeared twice in the eighth ancestral generation of No. 72316 and eight times in the sixth ancestral generation of No. 76002. Another pair consisting of female No. 43463 (dilute buff) and male No. 43398 (wild type) of the first laboratory-bred generation, appear in the fifth ancestral generation of No. 72316 and also in the sixth and seventh, respectively, ancestral generations of No. 76002.

Mouse No. 82106, which exhibited a different type of neoplasm, also had some of the same ancestors as did Nos. 72316 and 76002. In addition, female No. 59521 (*blandus,* Tularosa, black-tipped gray, light selection line) and male No. 58775 (hybrid between *Peromyscus maniculatus bairdii, P. polionotus albifrons,* and *P. leucocephalus*) were common ancestors to this mouse in the fifth ancestral generation and to No. 72316 in the third ancestral generation.

Since these mice had been exposed during their whole lives to essentially the same environmental conditions and since they have certain ancestors in common, it is possible that the development of the tumors could be due in part to hereditary factors. The evidence presented, however, does not prove that heredity is involved. If these neoplasms do have an hereditary basis, it is possible that the two types of neoplasms described could be produced by different hereditary combinations.

As pointed out above, no indication of a tumor was found in the records of the other relatives of the affected mice. No special examinations or autopsies, however, were made of most of these relatives. Only about 10 per cent of the relatives of all three affected mice were kept alive until they were over two years old. In 18 per cent of the cases the age at death or at disposal is unknown. The earliest known age of appearance of a neoplasm in the mice here described was two years. Thus, the majority of the relatives of these mice did not attain an age at which a tumor might be expected to develop. The failure of the relatives of these affected mice to develop tumors does not demonstrate an absence of hereditary tendency toward the neoplasms.

SUMMARY

Two cases of fibrosarcoma and one of an "ill defined, malignant neoplasm of glandular and connective tissue with caseation" are described from *Peromyscus* of the *maniculatus* group in the stocks of the Laboratory of Vertebrate Biology. The neoplasms occurred in mice all of which were more than two years of age, an age rarely attained in the wild state. Some inbreeding occurred in the strains represented by the mice, but no one of the individuals was highly inbred. All three affected mice are remotely related. It is possible, therefore, but is not proved that the development of the tumors might have been caused, at least in part, by hereditary factors.

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